THE CULTURING OF *JALMENUS EVAGORAS EVAGORAS* (DONOVAN) AND ITS ATTENDANT ANT, *IRIDOMYRMEX ANCEPS* (ROGER)

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Abstract

Techniques for maintaining an on-going laboratory colony of the myrmecophilous lycaenid, *Jalmenus evagoras*, and its attendant ant, *Iridomyrmex anceps*, are presented and discussed.

Introduction

In recent years there has been something of a renaissance in the study of the relationships between lycaenid larvae and pupae and ants. This is, in part, a consequence of the somewhat provocative paper of Malicky (1970) on the advantages and disadvantages of the association to both ant and butterfly. In addition, the new theoretical directions in the study of mutualism sketched, initially, by May (1976) have turned the attention of the population ecologist as well as the lepidopterist to these fascinating interactions.

For the size of our lycaenid fauna, Australia is particularly well-endowed with myrmecophilous or presumed myrmecophilous species (Common and Waterhouse, 1972; Kitching, in press). With all of these considerations in mind, we began in 1978, a detailed study of the morphology, behaviour and population dynamics of the common wattle-feeding lycaenid, *Jalmenus evagoras evagoras* (Donovan) and its attendant ant, *Iridomyrmex anceps* (Roger) basing our field work on colonies of butterfly and ant occurring at Mt. Nebo near Brisbane, Queensland.

In order to make a detailed investigation of the biology of any species of insect it is almost essential to be able to maintain cultures of the species in the laboratory so that all stages are accessible and of known history when required for experimental work. Although, as most lepidopterists are aware, *J. evagoras* is easy to rear from larvae to adult, to complete the cycle and produce generation after generation in the presence of the ant, is far less straight forward. We have developed techniques for culturing the pair of species and present them here in the hope that this may lead others to establish such cultures. The two species involved are sufficiently common to have potential both as teaching and research material, illustrating as they do a most dramatic and interesting natural interaction.

The insectary

We maintain our ants, butterflies and foodplants in a 4 m x 4 m x 2 m shade-house constructed of steel piping with a translucent fibreglass roof and a cement floor. The walls are of 30% shade cloth stapled to the inside of a timber substructure tied to the metal frame. A shade cloth curtain also screens the outer door. We subdivided the shade-house into a inner 1.5 m x 4 m x 2 m culture room and an adjacent service area and our butterflies fly free within this inner area (see below). The shade-house is provided with low (0.5 m high) plant benches of angle iron and stout weldmesh construction and, of those on which

ants are maintained, the bench legs sit in pots containing oil or detergent t prevent egress of the ants. Fig. 1 is a view of the culture room inside ou insectary.

Food plants

Larvae of *Jalmenus evagoras* feed on the foliage of a variety of species o *Acacia* appearing to prefer the compound-leaved varieties, although they are by no means restricted to them. In the Brisbane region the principal food plan is *A. irrorata* Sieb. ex Spreng. and it is this species and *A. decurrens* Willd. tha we have used in our cultures. Plants of *A. irrorata* were collected during th winter when 5-10 cm tall and kept in the shade-house for use in the ensuin summer season. We note in the field that butterfly larvae are absent from bushe greater than 2 m in height, and, in fact, have discarded plants in our culture when they exceed 1.5 m. On occasion, we have used nursery-bred *A. decurren* as substitutes for field-collected *A. irrorata* and the larvae and ants have thrive equally well on this species.

To restrict ants to particular plants we place plant pots on rubber stoppers in trays of oil as shown in Fig. 2, making sure the base of the pot is abov the level of the oil. We have found that ants will infest the soil of a pot if allowed contact with it, abandoning the plastic formicaria provided (see below). This can be prevented by stretching plastic sheeting or nylon mesh over th top of the pot and stapling, tying or sticking it in position, although the use of nylon is, at best, a temporary expedient as the ants will gnaw their way through it within a week.

The ants

The primary attendant of *Jalmenus evagoras* is *Iridomyrmex anceps*. This ant is highly polydomous and the main brood chamber is usually more than 20 cm below the soil surface. We attempted to remove nests whole, by digging them out and transferring them in large plastic bags. It proved difficult to obtain queens; only one excavation out of about ten was successful, this nest being in loose soil on a steep slope. The species is apparently polygynous as our most successful excavation gave us an entire nest in which there were three queens.

The collected nest can be kept for at least several days in its bag in a cool room. We tried manual sorting of cooled ants but this method is unnecessarily tedious and inefficient. As an alternative, we spread the cooled nest out over a large table covered in plastic and surrounded by a plastic strip coated with Tanglefoot®. Several formicaria (see below) are moistened liberally, covered with aluminium foil with one entrance left open and left at various locations on the table. The ants, when they warmed up, regroup themselves and collect their scattered brood into the moist, dark formicaria. After one day, the formicaria could be closed up and the sub-colonies so obtained, removed.

Our standard culture uses formicaria of a design provided by Dr R. W. Taylor. These comprise a perspex dish constructed with a central hole in its base which is filled with plaster of Paris. The sides of the dish are pierced for three (or more) exit tubes which can be stoppered or connected with other

Figs 1, 2. (1) view of the *Jalmenus* rearing room showing food plants and oil trays; (2) a single food plant equipped with ant excluder (above soil) and formicarium adpressed to the stem. Pupal debris and branches stripped by feeding larvae can be seen.

units. A tight-fitting lid completes the unit. The formicaria are maintained on a moistened pad (filter paper will suffice) so that the plaster of Paris remains moist. They are covered with metal foil and kept in an open plastic lunch box, the sides of which are coated with the anti-friction lacquer Fluon®. The formicaria are equipped with about a tablespoon of soil to allow the ants to have some control over nest humidity. Originally the colonies were fed on honey solution and dead spiders, however, we now use a totally liquid diet of 5: 8 honey: water (effectively 50% sugars) mixed 2 : 1 with egg yolk (50% protein, 20% fat). A small colony (50 workers and 20 larvae) will go through about 0.1 ml/day during alate production. The feeder solution is administered in small 10 mm lengths of 5 mm plastic tube, which are filled from a syringe. Clear tubing allows inspection of amounts of food remaining. The tubes are discarded after use.

Adult ants will last the winter without feeding. One small queenless colony in fact persisted with 47 adults, 12 pupae and 19 larvae from April to September unfed—they may however have eaten away at a larger initial brood or derived nutrition from small organisms in the soil which was included in their formicarium.

Handling, transferring and mixing the colonies is best achieved by anaesthetizing them with CO_2. This keeps the adults inactive for up to a minute. These ants are prone to nest shifting, and brood will be transported by workers on the slightest disturbance. As already mentioned, given the choice of soil or formicarium, the colony will choose soil. To avoid this, the soil surface in pots must be covered to prevent invasion unless having the colony in the pot presents no disadvantage.

Ants were allowed access to the food plants by placing a formicarium with its only exit adpressed against the base of the shrub (Fig. 2).

The butterflies

The laboratory colony was established by bringing in from the field cut-stems of the food plant together with attached clusters of larvae and pupae. These stems were tied against branches of the potted wattles in the culture room and the attached larvae transferred themselves to the living foliage within twelve hours. These immature stages, tended by the ants which had access to the trees, produced the first generation of adults. In addition, overwintering egg batches have been brought in on occasion, incubated, and the hatchlings transferred to host trees.

Adults will mate shortly after emergence even in restricted spaces (initially we had them in 0.5 x 0.5 x 1.0 m gauze cages) but we obtained no oviposition in such confinement. Only when we established our cultures, free-flying, in the much larger culture room did oviposition follow mating. In the field, on the more weathered wild host shrubs the egg-laying females usually seek out crevices in the bark in which to lay their batches of up to 20-30 eggs. In the case of our cultured shrubs, which had smooth, undamaged bark for the most part, we found that suitable crevices for oviposition could be manufactured either by making slits in the bark with a scalpel or, more conveniently, by binding small sections of the stem with a rough, fibrous string. The butterflies

accepted this latter expedient readily as an alternative site for oviposition. It has been our impression also that oviposition was more likely on those shrubs which still had either living immature stages on them or the debris from such stages in the form of larval exuviae or pupal skins. We are currently carrying out choice-experiments to determine the relative attractiveness of trees with and without pupal debris and/or ants and the results of these will be reported in due course.

Adult feeders were provided in the culture room and were made from ranks of four 5 mm glass tubes mounted vertically in wooden blocks, each surmounted by a plastic corolla (culled from commercially available plastic flowers) and a small cube of sponge. The tubes are maintained full of 50% aqueous hoeny solution which is absorbed by the sponge and from which the butterflies feed readily. Butterflies will feed from petri dishes containing cotton wool pads soaked in honey solution but the feeders described seem to provide a more accessible and reliable food source for them.

General comments

We maintained the culture of *J. evagoras* and its attendant ants in the manner described above throughout the summer of 1978-79 during which period it provided ample material for experimental and morphological work. At the end of the summer season the adults laid diapausing eggs and, in this fashion, the colony survived over the winter, the overwintering eggs hatching and seeding the following summer's colony. The culture is still in good health and we feel confident it can be so maintained. Periodically we have brought in further material from the field to obviate any problems of low vigour in the laboratory arising from inbreeding.

The species involved present many opportunities for work on various aspects of the butterfly, the ant and the interaction between the two. We commend them both as research and teaching tools.

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